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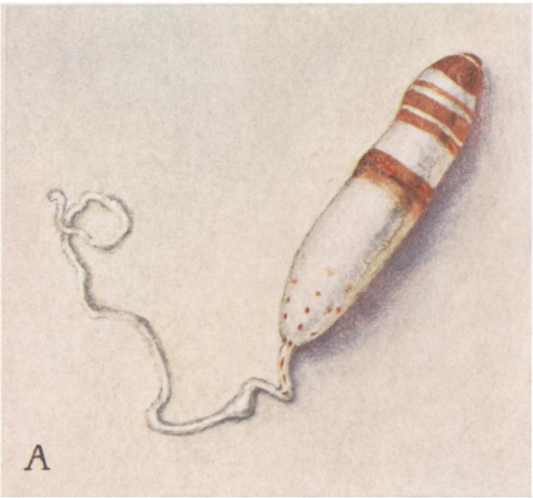

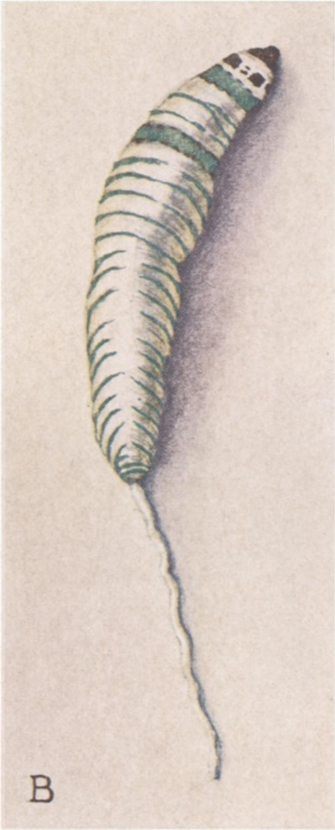

PLATE VIII

EXPLANATION OF PLATE VIII

Fig. A. The mature sporocyst of *Leucochloridium problematicum.*

Fig. B. The mature sporocyst of *Leucochloridium macrostomum*, after Carus.

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LEUCOCHLORIDIUM PROBLEMATICUM N. SP.*

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While examining snails at Fairport, Iowa, in the study of *Lissorchis fairporti* Magath, during the summer of 1917, two snails were found which harbored mature sporocysts of a parasitic trematode of the genus Leucochloridium, and which is to be designated as *Leucochloridium problematicum* n. sp.

The first snail was obtained from Experimental Pond 2 D and was a species of *Succinea retusa* Lea, while the second was a *Planorbis trivolvis* Say, from Experimental Pond 5 D. On further study seven other snails, all of the latter species, were found to be infected with younger sporocysts which could be demonstrated by careful dissection and examination of the livers. In all, records of 209 snails were made, including the following species and numbers of each:

Planorbis trivolvis	120
Succinea retusa	48
Lymnaea obtussa exigua	23
Physa heterostropha	18

This gives an infection of 4.5 per cent. for all; or 2 per cent. for *Succinea retusa* and 7 per cent. for *Planorbis trivolvis.*

In examining the literature for reports of Leucochloridium from America, only two references can be found, and both of them are open to question. The first is that of Call (1898), who remarks under the description of *Succinea obliqua* in Indiana, "the tentacles are rather large and thick, club-shaped, and are often the home of a stage in the development of a planarian."

The second account is given by Ward (1917), who states that Mr. Bryant Walker, in a personal letter, reported finding the larval stage of a Leucochloridium in *Succinea ovalis* in Michigan. It is of course impossible to say whether or not these reports refer to the species or even the genus herein described, but it is possible that since both reports come from nearby states the same worm was found.

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* Contribution from the Laboratory of the United States Bureau of Fisheries, Fairport, Iowa.

Figures 3, 4 and 5 were drawn by Mr. C. W. Shepard, artist in the Department of Anatomy, University of Illinois, College of Medicine, Chicago, Ill.

Leucochloridium is a genus created by Carus in 1835 to contain a species of larval trematode which he named *Leucochloridium paradoxum*. However, Zeller (1874) showed that this larval form developed into the adult trematode that Rudolphi found in the cloaca of *Motacillae luscinae* in 1803. This early parasitologist called the worm *Fasciola macrostoma,* but in 1809, recognizing its difference from the liver fluke of the sheep, changed the genus name to Distomum, designating the worm as *D. macrostomum*. Monticelli (1888) thought that the description of *D. macrostomum* was sufficient to justify creating a new genus to contain it and called the genus Urogonimus naming *U. macrostomus* as the type. He gave practically no description of the genus except to call attention to the fact that the genital pore was posterior. In 1893 he described *U. ceratus* as a new species. Braun (1896) used in his text both Leucochloridium and Distomum in referring to the species of Carus, but apparently accepted the work of Monticelli, and the genus Urogonimus. Looss (1899) also accepted the genus Urogonimus and described a new species, which he called *U. insignis*. At this time he established a new genus to contain *Urogonimus rossittensis* (Mühling). The two genera were then placed in the subfamily Urogoniminae, with Urogonimus as the type genus.

Stiles and Hassall (1898) stated that they were inclined to adopt *Urogonimus* as a genus in view of the fact that the law of priority did not entitle larval genera to be used as names for adults.

It remained for Poche (1907) to make this final step, and he stated that the genus should be known as Leucochloridium, with *L. macrostomum* (Rud.) as the type, which necessitated the substitution of Leucochloridiinae for Urogoniminae of Looss. This of course does away with the original specific name of Carus for the larval form.

The decision of Poche has been accepted by Lühe (1909) who places the genus in the superfamily Distomata Retizus as Group IV, forms with genital opening posteriorly, and gives a clear cut generic description. He states that the only species found in Germany is *L. macrostomum,* the adult of which has been found in *Rallus aquaticus L., Gallinula chloropus* (L.), and *Ortygometra porzana* (L.). The larval form he reports from *Succinea putris* L. Ward (1917) also accepts this disposition of the genus, and it seems to the author to be the best that can be made, since the genus was clearly defined under the name Leucochloridium and could be so recognized.

Apparently the only larval form that has been reported is that of Carus, who first found it in *Succinea amphibia* in 1833 (reported in 1835) on an island in the Elbe. He states that a friend of his found a similar sporocyst in a snail in Halle in 1825, and it has been reported from Leipzig by Heckert. The description of Carus is of course, not complete, and two of his figures are reproduced in the present paper.

He found a snail whose tentacles seemed to be stained green and white; on close study these were seen to be two bodies, not unlike the larvae of certain insects, which could be retracted into the body of the snail or extended into the tentacles. The mass pulsated and was white with green bands all round it, and brown "warts" at the tip. Near the tip several bands were wider than the others. At the proximal end of the green and white body was a thread-like structure which he later traced into the liver of the snail, where were found numerous small knob-like bodies connected to it by other threads. He tore one of the larger sacs open and found it contained about 300 small worms, each in a little sac of its own. On the inside of the sporocyst were little bodies or cells which he said developed into the worms. These were, according to him, one-sixth of a line long and had well developed organs, which he did not describe in a very complete manner. He further called attention to the fact that the individual sacs of the worms had connections with the two suckers. In discussion of his findings he quotes the following interesting note from Rudolphi (1809): "in tentaculis *Helicis putris L.* Augustus Ahrens Halae Septembri corpuscula reperit, quae omnino huc facere et novum genus sistere videntur."

Following this many observations were made upon this worm, by Leuckart, von Siebold and others, until finally Zeller (1874) showed the relation between the larval form in the snail and the adult worm in the cloaca of singing birds, found many years before by Rudolphi.

What Zeller described in brief Heckert (1889) enlarged upon and gave a very extended account of the histology, morphology and life history of the species. He found the larvae not rarely in the snails near Leipzig. The ramifying tubules penetrated the liver of the host; these tubules were filled with a serous fluid and germ-spheres from which the larvae developed. Parts of the tubules extend up into the tentacles, and these portions are highly colored. The tubules have three muscular layers, a longitudinal, a circular and a diagonal layer. Below this dermo-muscular layer is bright green pigment arranged circularly. The sporocyst has the same structure as the tubules. There is an exterior cuticula, a dermo-muscular tube, and inside this a layer of cells which vary in size with the stage of development. Still inside of this is a membrane with distinct cellular elements, the cells of which differentiate and drop off, eventually forming the larvae. The order of development of the organs of the larvae is as follows: genital cells, suckers, pharynx, enteron, excretory system and nervous system.

There is a double ecdysis, but the cuticula remains with the larva, forming a protective covering, with fluid between it and the worm and the suckers connected to the covering. The Sylviidae are the true hosts and the adult is found in the cloaca; eggs are formed at the end of eight days. The shells are thick, $1/30$ mm. long and at the hinder end of a ciliated comb, there is a powerful cone which acts as a steering organ. When the eggs were fed to Succinea after eight days they were found in the liver; they bored through the stomach wall with their head cones. Heckert further noted that in the adult the genital pore is terminal or dorsal and not ventral.

Zeller (1874) says the larvae are white, the body flat and broader at the anterior end than posteriorly. The integument is thickly covered with fine hairs or cilia. The anterior end of the body is very peculiarly constructed with its collar-like projection of the entire integument, which overlaps the mouth sucker and sticks up high behind. There are great numbers of unicellular glands in this projection. The suckers are large and very powerful, the mouth sucker being larger than the ventral one. The digestive apparatus consists of a powerful pharynx, a very short esophagus and the intestinal crura which describe a graceful arc as they extend to the posterior part of the body. The excretory system has a short contractile terminal bladder. The rest of the system consists of small tubules which seem to end blindly as fine branches.

The genital organs are quite well developed, and both the male and the female organs lie in the posterior part of the body. The two testes are more or less egg-shaped. One lies immediately posterior to the ventral sucker on the right, the other further back and on the left side of the body. The duct from the first leads out towards the other testis. Both ducts join together and the vas deferens passes posteriorly, lying in the posterior part of the body in a spherical cirrus sac, ending in an extensible cirrus at the posterior end of the body. The cirrus is not easily demonstrated in the larvae, although it is prominent in the adult.

The ovary lies between the two testes, nearer the posterior one. It is round in shape and has a short duct leading from it. This receives first the duct by which the spermatozoa enter from the vagina which opens on the dorsal surface anterior to the excretory opening. After this the duct bends and receives from beneath, at right angles, the ducts from the yolk glands. In the larvae these ducts enter the oviduct in a "rounded-body" which was thought to be the shell gland. The oviduct continues as the true uterus, which at first passes to the left and anteriorly to nearly the pharynx, there crosses to the right above the ventral sucker and descends to the posterior part of the body on the right side where it opens near that of the cirrus sac. The uterus is empty in the larvae, but in the adult the eggs are dark brown and measure 0.025 mm. by 0.014 mm.

The description of the adult *L. macrostonum* does not concern us here, but one might state that the adult structure is accurately forecasted by the larvae, and that in the ceca of the singing birds they reach a maximum length of 1.8 mm. by 0.80 mm.

However, the description of *L. insignis* is important for us, and following is given in brief Looss' (1899) description of the species. The worms were found in the cloaca of *Fulica atra.* They are about 3 mm. long and 1.35 mm. broad. The anterior end is thicker and more rounded, the posterior flattened and also somewhat rounded. The suckers are very powerful and large; the mouth sucker has an elevated margin around it. This sucker (0.73 mm.) is as large as, and perhaps larger than, the ventral one. The skin is not ciliated. The mouth leads into a very short prepharynx, and this into a powerful muscular pharynx which is 0.3 mm. in cross section. The esophagus follows immediately. The intestinal crura pass back as far as the genital pore. The excretory pore lies on the dorsal surface 0.27 mm. from the posterior tip. The genital pore is on the dorsal surface 0.17 mm. below the excretory pore. The structure of the genital organs is characteristic for the genus Leucochloridium. The cirrus sac is bulbiform or pear shaped, 0.33 mm. by 0.13 mm. It is a connective tissue mass through which passes the ductus ejaculatorius and from which projects the cirrus; the latter is large in cross section and with thick walls. The pars prostatica is short with but few prostatic glands. The tube decreases and, lying free in the parenchyma is found the thick, muscular-walled seminal vesicle. Into this pass the two ducts from the testes which lie on either side of the body, one behind the other. Between them is the small ovary, lying close to the posterior testes. A receptaculum seminis is lacking. Laurer's canal passes posteriorly and dorsalward to end in the excretory vesicle. The yolk glands lie on the side of the body and stretch from the posterior end of the intestinal crura to the level of the pharynx. The uterus follows the general form of the type species. The egg shell is thick, but not very dark colored, measuring 27μ by 15μ.

Lühe (1909) has defined the genus Leucochloridium as follows: Small distomes, thickened at the rounded end, oval in cross section, with well muscled bodies. Large, very strong suckers. Skin ciliated or smooth. Pharynx strong, esophagus very short. Intestinal ceca very thin and reaching to the posterior end. Excretory pore a little distance from the posterior tip on the dorsal surface. Excretory bladder simple, short. Genital opening at the end. Cirrus sac posterior but enclosing the cirrus and ductus ejaculatorius only. The short prostatic part is also muscular, a spindle shaped seminal vesicle lying free in the parenchyma, passes laterally and obliquely to the posterior part of the body from the testes; the ovary lies between the testes. A receptaculum seminis is lacking. Laurer's canal is present. The yolk glands are numerous, and placed at the sides of the body, lateral to the intestinal ceca. The uterus has numerous coils, entirely encircling the sucker (ventral). Eggs numerous, small with thick shells. Habitat: the cloaca of birds.

LEUCOCHLORIDIUM PROBLEMATICUM

The description of the sporocyst was made from two mature sporocysts which were essentially alike in all respects. The sporocyst of this species is 1.4 cm. long by 0.33 cm. wide and pointed at both ends. The proximal end is continued in a thread-like tube which a little distance away has in its course a small fusiform swelling. The tube then continues into the liver of the snail where many small knob-like projections appear in the hepatic tissue and are connected by thread-like processes. The wall of the sporocyst is rather thick and tough, being 6μ thick. It is white and translucent. The distal half of the sporocyst is banded with deep golden red bands which show very accurately in figure A (Plate). Some of these are darker than others and the lighter ones tend to be more yellow. No bands appear beyond the distal half. The proximal ¼ is flecked with bronze spots, minute in size, but readily seen. These flecks are also present for a short distance on the tube projection. By comparison with the sporocyst of *L. macrostomum,* it will be seen that this sporocyst is essentially different in marking and color. They are both near the same size.

A cross section of the wall of the sporocyst is seen in figure 14. It is made up of an outer longitudinal layer and an inner circular layer of muscles; the former becomes divided by diagonal fibers which serve to break them up into bundles. The nuclei are irregular and branching. Pigment is present in the cells beneath the outer layer. The sporocyst is capable of pulsation, when mature projects from the snail's tentacle, and each contains about 100 larvae which are very active when freed in water; each has a little sac covering it which is quite transparent and has connections with the two suckers. The sac in the living material was 2.6 mm. by 1.4 mm.

The larvae themselves are about 2.2 mm. long and 0.85 mm. wide. They are quite active and very transparent. The cuticula is beset with fine cilia and some of the organ systems are best studied from living material. This form has two well developed suckers which are readily seen with a low power lens. The oral sucker is the larger of the two, and has its opening near the anterior end of the body. This sucker is 0.24 mm. wide and 0.4 mm. deep. Its posterior end leads into a rather powerful muscular pharynx which is 0.17 mm. by 1.15 mm. From this the lateral intestinal crura arise and each one passes down on either side of the worm to the level of the opening of the excretory canal. These crura are not especially narrow as in *L. macrostomum,* but are in *L. problematicum* 0.55 mm. in diameter. The ventral sucker is 0.34 mm. in cross section, is circular and situated about in the middle of the antero-posterior axis.

The excretory system is not unlike that of *L. macrostomum,* and consists of a rather simple set of tubules. The flame cells are situated throughout the body and seemed to be collected in a larger pair of tubules, one right and one left. These pass down in the body parenchyma on the ventral side of and median to the intestinal crurae to within 0.10 mm. of the end of the crura. Making a rather sharp turn each tubule, expanding in diameter, passes anteriorly and laterally to the crura to within 0.2 mm. of the anterior tip of the body. Another sharp turn here occurs and with increasing diameter each tubule passes posteriorly between the ascending ramus and its corresponding one of the intestinal crura gradually towards the mid line, where it joins its fellow, after a slight fusiform enlargement, in the mid line 0.09 mm. from the posterior tip on the dorsal side of the body. Almost immediately the excretory pore opens to the exterior. These posterior enlargements of the descending rami are pulsating in character, filling and emptying every few seconds.

The genital organs are quite well developed in these larvae and one has no difficulty in outlining them, as they are certain to occur in the adult. The testes are two in number, one anterior lying to the right of the mid line, the other posterior, 0.13 mm. behind the former and to the left of the mid line. There passes from each and towards the other a small duct, the two joining 0.03 mm. from the posterior testis; from this union there arises a small duct which passes posteriorly, slightly to the right of the mid line, through the cirrus sac and opens as the ductus ejaculatorius to the exterior 0.58 mm. from the posterior tip in common with the uterus. The cirrus sac is fusiform, tapering more sharply anteriorly and is 0.16 mm. by 0.07 mm. No cirrus is developed in this stage of the larvae.

The ovary is spherical in shape, 0.052 mm. in diameter and lies on the left side of the body, at a level between the two testes and nearer the posterior one. There passes from it towards the mid line a short oviduct which almost immediately is joined by Laurer's canal. This canal passes posteriorly and ends in the excretory duct immediately before it opens to the exterior, just as described in the case of *L. insignis* by Looss. The oviduct after a short bend receives the two ducts from the embryonic yolk glands. Following this the oviduct makes a twist upon itself and then passes anteriorly as the ascending uterine branch, first in the mid line, then to the left of the ventral sucker. This branch turns toward the mid line anterior to the sucker and passes posteriorly to the right, then in the mid line and by a more or less straight course to the genital pore on the dorsal surface of the body.

The oviduct receives the yolk gland ducts, which are in reality the shell glands, and makes a coil within an organ called the "round body." This is perhaps a gland which acts in some way upon the shell gland substance, perhaps as a precipitin, and was noted in the larva of *L. macrostomum*, but not in the adult.

Several unicellular glands are found just posterior to the ventral sucker and also along the anterior margin of the oral sucker. Their function is one of speculation, and no data is at hand to make even a profitable explanation for their presence.

TABLE OF MEASUREMENTS

	L. macrostomum Larva	L. macrostomum Adult	L. problematicum Larva	L. insignis Adult	L. cercatum Adult
Length	0.8	1.8	2.2	3.0	4.0
Width	0.45	0.9	0.85	1.35	1.2
Anterior sucker	0.17	0.20	0.39	0.73	0.60
Ventral sucker	0.14	0.20	0.34	0.69	0.72
Width pharynx	0.075	0.16	0.15	0.30	0.22
Testes					
Anterior	0.058	0.20	0.074	0.22	0.26
Posterior	0.060	0.22	0.061	0.20	0.26
Ovary	0.059	0.20	0.052	0.13	0.24
Cirrus sac	0.061 by 0.061	0.15 by 0.16	0.07 by 0.16	0.13 by 0.33	0.13 by 0.34
Round body	0.041		0.043		
Larval sac	1.1 by 0.8		2.6 by 1.3		
Sporocyst	1.7 by 0.25 cm.		1.3 by 0.3 cm.		

All measurements in millimeters.

DISCUSSION

In looking over the description given by Looss of *L. insignis* one is struck with the fact that it is very much like the larval form described in this paper, and the question at once arises as to whether this is in reality the larval form of Looss' species. *Fulica atra* is not found in Fairport, but the corresponding American species is, and it is not impossible that these two trematodes are one and the same. The author, however, does not feel warranted in coming to this conclusion chiefly from a geographical reason, although there is nothing definitely present or absent in the larvae to distinguish it from *L. insignis*. I have, therefore, given it a name symbolic of my conception of the species, and it remains yet to find or experimentally develop adults of *L. problematicum*, which may be compared with *L. insignis*.

In addition to this rather perplexing situation one cannot help but note the similarity between the species of Looss and that of Monticelli. Unfortunately, the description of the latter author is not very complete, and if it is correct and the figure is accurate the form may not even be a member of this genus, because, according to him, the ovary is posterior to both testes, while the genus Leucochloridium is characterized by the location of the ovary between the two testes. However, it seems to me that Monticelli has made an incorrect observation on this point, and the organ labeled "posterior testis" is really the ovary. Granting this, the two forms are then very closely related and come from the same bird genus. The general size of the worms and their organs are very nearly alike, and the shape of the cirrus sacs are also similar. In the absence of detailed information concerning the species and the fact that Looss, being cognizant of the description of Monticelli, considered his different after a study of his own material, makes it hazardous for me to say that these two worms are the same.

It is not possible to say absolutely, therefore, that the larval form *L. problematicum* is or is not the larval form of *L. ceratum* or of *L. insignis,* or still yet whether it is or is not the larval form of both, they being the same species.

CONCLUSIONS AND SUMMARY

1. A larval trematode, from sporocysts found in *Succinea retusa* and *Planorbis trivolvis,* has been described from Fairport, Iowa.

2. This trematode belongs to the genus Leucochloridium, but is unlike the only larval form of this genus ever described. It has been named *Leucochloridium problematicum.*

3. This larva is remarkably like the adult *L. insignis* (Looss) and *L. ceratum* (Monticelli), and it is possible that they are one and the same species.

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ABBREVIATIONS USED IN PLATES

a t	anterior testis
a s	anterior sucker
a ut	ascending uterus
c	cirrus
c m	circular muscle
c s	cirrus sac
d ut	descending uterus
e d	excretory duct
e p	excretory pore
g p	genital pore
i	intestinal crura
l c	Laurer's canal
l m	longitudinal muscle
o m	oblique muscle
ov	ovary
ovd	oviduct
p	pharynx
p t	posterior testis
r	rounded body
u g	unicellular glands
u t	uterus
v d	vas deferens
v dt	vitillarian ducts
v s	ventral sucker
y g	yolk glands

The magnification line by the side of each drawing represents a length of 0.2 mm. in all except Figures 13 and 14, in which it is 0.05 mm. long, and Figures 12 and 23, in which it is 0.1 mm. long.

EXPLANATION OF PLATE IX

Fig. 1.—Toto drawing of *L. problematicum* from the ventral aspect.

Fig. 2.—Drawing of the reproductive organs of *L. problematicum* from the dorsal aspect. × 150.

Fig. 3.—Toto drawing of the larva of *L. macrostomum* after Zeller. × 120.

Fig. 4.—Toto drawing of the larva of *L. macrostomum* after Carus.

Fig. 5.—Toto drawing of the adult of *L. macrostomum* after Zeller. × 60.

Fig. 6.—A group of young sporocysts of *L. problematicum* dissected from the liver of *Planorbis trivolvis*. × 8.

Fig. 7.—Longitudinal and transverse sections through *L. problematicum*. Longitudinal sagittal section.

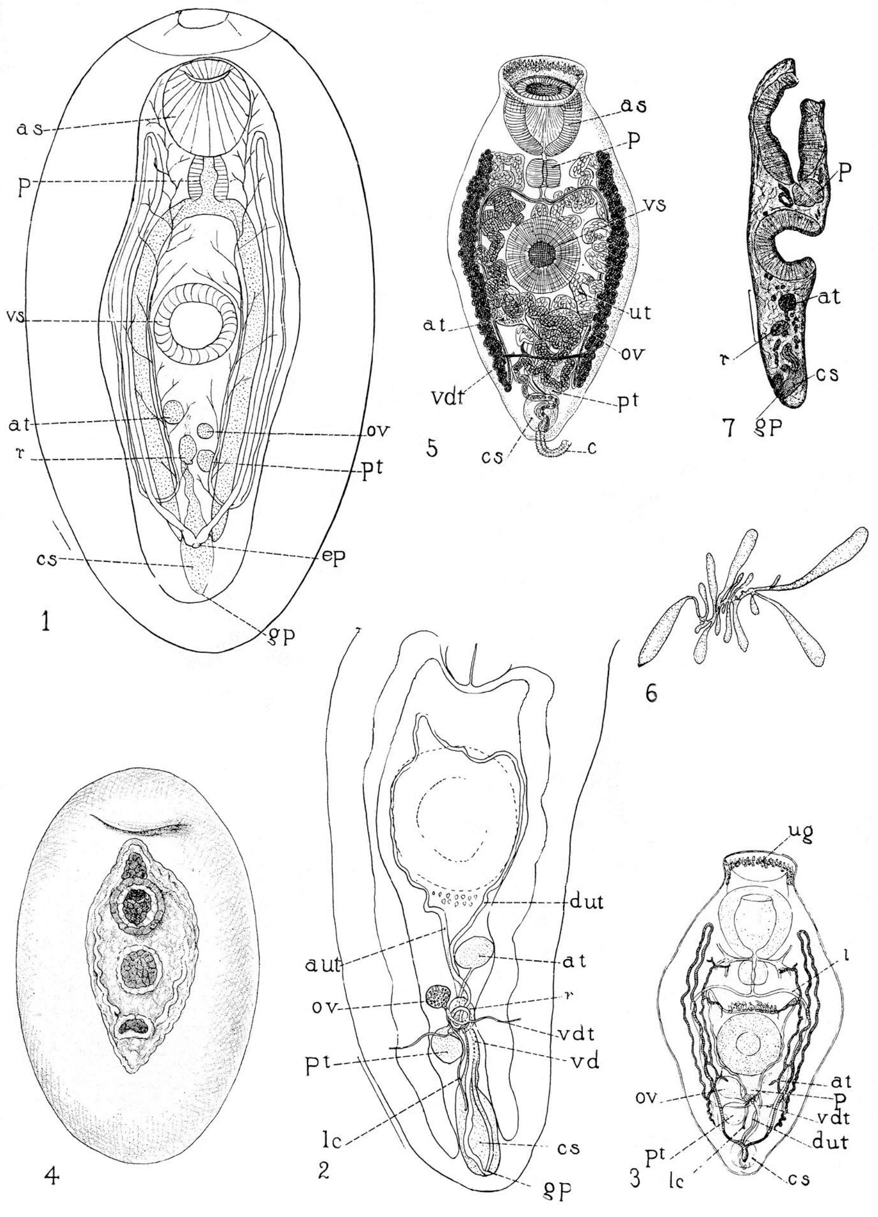
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PLATE IX

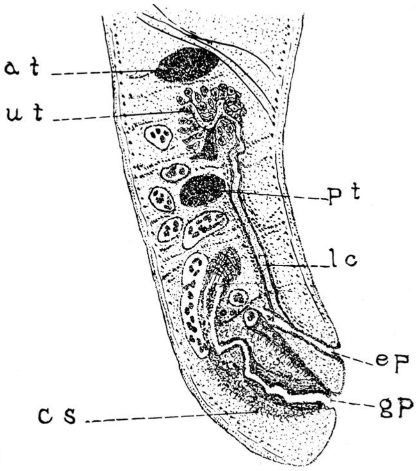
EXPLANATION OF PLATE X

Fig. 8.—Toto drawing of *L. insignis* after Looss. × 30.

Fig. 9.—Longitudinal sagittal section through the posterior end of the body of *L. insignis,* after Looss.

Fig. 10.—Toto drawing of *L. ceratum* after Monticelli.

Fig. 11.—Posterior region of *L. ceratum,* after Monticelli.



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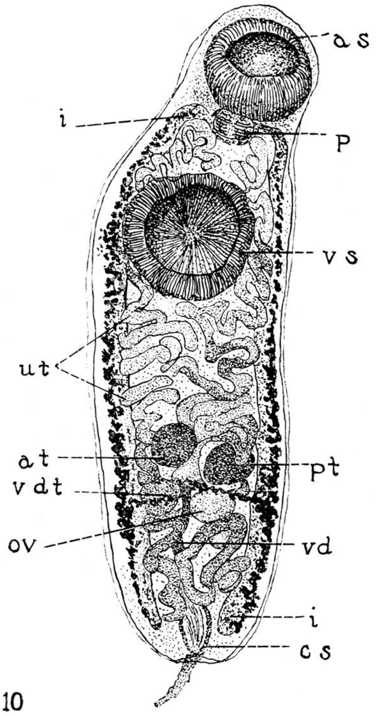

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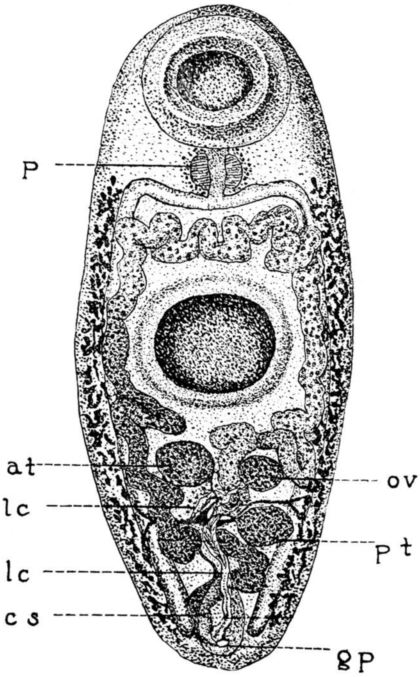

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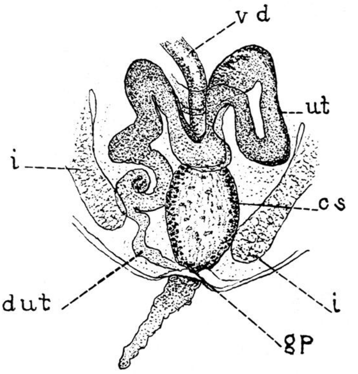

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PLATE X

EXPLANATION OF PLATE XI

Leucochloridium problematicum

Fig. 12.—Longitudinal sagittal section through the anterior margin.

Fig. 13.—Longitudinal sagittal section through the posterior end of the body.

Fig. 14.—Transverse section through the sporocyst wall.

Fig. 15.—Transverse section through the anterior sucker.

Fig. 16.—Transverse section through the anterior sucker, posterior to Figure 15.

Fig. 17.—Transverse section through the ventral sucker.

Fig. 18.—Transverse section through the pharynx.

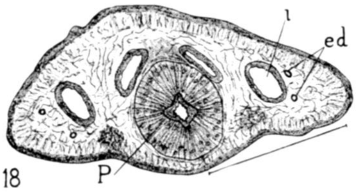
Fig. 19.—Transverse section through the anterior testis.

Fig. 20.—Transverse section through the ovary.

Fig. 21.—Transverse section through the rounded body.

Fig. 22.—Transverse section through the posterior testis.

Fig. 23.—Transverse section through the cirrus sac.



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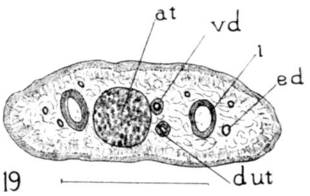

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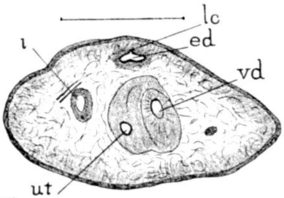

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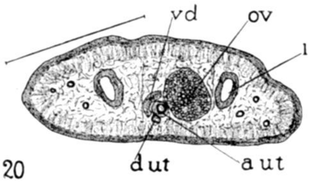

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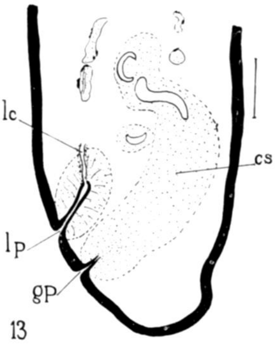

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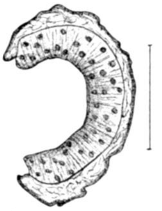

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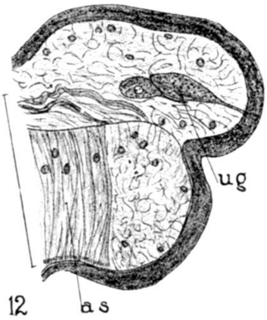

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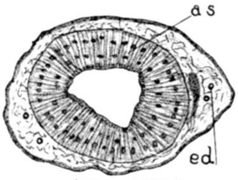

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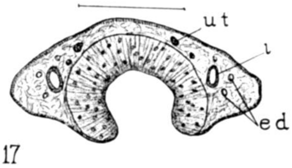

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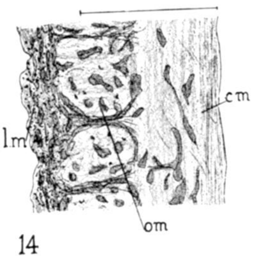

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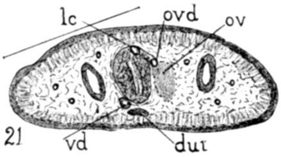

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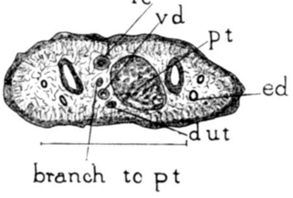

22

PLATE XI